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REPORT OF THE GOVERNMENT-INDUSTRY MEETING TO DISCUSS A NEWFOUNDLAND PROGRAM ON NON-MATURING SALMONIDS FOR LOCAL AQUACULTURE PERFORMANCE EVALUATION

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ATLANTIC FISHERIES
ADJUSTMENT PROGRAM

PROGRAMME D'ADAPTATION DES
PÊCHES DE L'ATLANTIQUE

Minister of Supply and Services Canada 1991
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EDITOR'S COMMENTS

This document is a result of an initiative by the Department of Fisheries and Oceans, Newfoundland Region, to examine and evaluate methods for the production of non-maturing salmonids in support of aquaculture development. The government-industry consultations that have resulted in the present report are in keeping with Science Sector Operational Priorities on scientific dialogue and improved communications with the scientific community and with the Department's clients.

In assembling this document, I have relied wholly on comments made during the government-industry meeting. In appreciation of the contribution of time and the opinions expressed by the participants, I have included the meeting minutes as an Appendix to this document. While these minutes are not verbatim, I believe they depict a very close representation of what was expressed at the meeting. Any errors or omissions are solely my own responsibility. The section of the report on "Highlights of Participant Discussions" is composed entirely of extracted passages of the minutes. Editing has been minimal and solely for the purpose of readability. As such, this document contains the opinions of the participants. Any statements or views presented here are totally those of the speakers. The Department of Fisheries and Oceans does not accept any responsibility for statements made by its clients.

This meeting was funded by the Development component of the Atlantic Fisheries Adjustment Program.

ABSTRACT

Pepper, V.A. [ed]. 1991. Report of the government-industry meeting to discuss a Newfoundland program on non-maturing salmonids for local aquaculture performance evaluation. Can. Ind. Rep. Fish. Aquat. Sci. No. 208. v + 26p.

This report documents interests of the Newfoundland salmonid farming industry regarding aquaculture brood stock and Department of Fisheries and Oceans (DFO) regulations. While some industry representatives conveyed both frustration and hostility regarding DFO legislation and policy, participants generally were in favour of a joint, cooperative venture with government, to implement a Newfoundland program on non-maturing salmonids. It was agreed that both levels of government would collaborate to provide support to performance evaluation of triploid salmonids in Newfoundland. A pressure vessel, as an essential prerequisite to all participants in a demonstration program, would be made available to industry as a whole. Pending identification of sufficient funding, a Newfoundland program on non-maturing salmonids would start in October, 1991.

RÉSUMÉ

Pepper, V.A. (éd.), 1991. Rapport de la réunion entre le gouvernement et les salmoniculteurs sur un programme d'aquiculture local portans sur l'évaluation du rendement des salmonidés qui n'atteignent pas la maturité. Can. Ind. Rep. Fish. Aquat. Sci. No. 208. v + 26p.

Le présent rapport rend compte de l'intérêt des salmoniculteurs de Terre-Neuve à l'égard des stocks géniteurs en aquiculture et des règlements du ministère des Pêches et des Océans. Malgré la frustration et l'hostilité manifestées par certains industriels à l'endroit des lois et des politiques du Ministère, la plupart des participants ont favorisé une approche conjointe et coopérative avec le gouvernement pour établir à Terre-Neuve un programme de salmonidés qui n'atteignent pas la maturité. Il a été convenu que les deux niveaux de gouvernement collaboreraient dans le but de soutenir une évaluation du rendement des salmonidés triploïdes à Terre-Neuve. Un navire sous pression, indispensable à un tel programme de démonstration au dire de tous les participants, devrait être mis à la disposition de l'industrie. Si les fonds nécessaires étaient alloués, un programme terre-neuvien de salmonidés qui n'atteignent pas la maturité serait amorcé dès octobre 1991.

INTRODUCTION

A meeting of industry representatives, Federal and Provincial government officials, and interested research scientists from Newfoundland took place on July 10, 1991. The meeting was convened at the Freshwater Resource Centre in St. John's, Newfoundland. Vern Pepper (Science Branch, Department of Fisheries and Oceans) assumed the duty of chairperson for this meeting.

Prior to the meeting, the chairperson distributed to the invited participants, copies of a report (Pepper 1991a) documenting the proceedings of a workshop on methods for the production of non-maturing salmonids held in Dartmouth, Nova Scotia in February, 1991. The report was intended as background to the present meeting.

The chairperson welcomed participants and explained that the meeting was a Newfoundland Region initiative, funded under the Development Component of the Atlantic Fisheries Adjustment Program (AFAP). He thanked the DFO Industry Development Division for providing the AFAP funding and for this opportunity to bring together representatives of the salmonid farming industry, as well as the academic community and interested government agencies. Participants in the July 10 meeting are identified in Appendix 1.

PURPOSE OF MEETING

The chairperson reviewed historic involvement of the Science Branch of DFO with the issue of Atlantic salmon brood stock development in Newfoundland and explained that DFO actions on this topic have been determined largely by mandate concerns as defined by the Newfoundland Fishery Regulations and discussed by Pepper (1991b). In light of growing concerns within the international scientific community about the potential consequences of interbreeding of wild salmon stocks and escapees from salmon cages, the Newfoundland Region of DFO has limited importation of salmon eggs into Newfoundland, by the Bay d'Espoir Salmon Growers Co-operative, to 100,000 eggs per year. A further constraint imposed by DFO on these importations is that industry was not to develop a brood stock from the progeny of imported eggs.

In its attempts to provide options for brood stock development, in support of Newfoundland salmonid aquaculture, Science Branch has been pursuing three separate initiatives as follows:

1) evaluation of local wild salmon stocks and their aquaculture performance characteristics;
2) identification of alternative breeding strategies and their relative potential to provide the greatest performance improvement benefits in the shortest time for the lowest cost; and,
3) demonstration of technology for suppression of salmonid maturation and evaluation of performance of reproductively incapacitated salmonids relative to normal, reproductively active salmonids.

The first of these initiatives has been ongoing for several years. Three Newfoundland salmon stocks (Grey River, Conne River and Exploits River) have been raised in the Bay d'Espoir Hatchery. All of these stocks demonstrated early maturation. A further attempt is in progress as a cooperative venture between Science Branch and SCB Fisheries of Bay d'Espoir. Grand Codroy (St. Georges Bay, Newfoundland) eggs were taken to the Bay d'Espoir Salmon Hatchery in 1989. Performance of the progeny of the Grand Codroy parents in the hatchery was similar to progeny of the St. John River stock. Grand Codroy smolts from the hatchery now are in sea cages in Roti Bay in a comparative performance experiment with smolts of the St. John River salmon stock.

Evaluation of alternative breeding strategies to develop a salmon brood stock for Newfoundland was undertaken under a contract (funded under the Canada-Newfoundland Inshore Fisheries Development Agreement) with Tri-Gen Fish Improvements Ltd. of British Columbia. This firm also designed a breeding program for the British Columbia salmon farming industry. A contract report (Peterson et al. 1990) is available

in the library at the Northwest Atlantic Fisheries Centre in St. John's. This report recommends a comprehensive program of "family tie" matings and overlapping generations (see Appendix 2 for explanation of technical terms).

One recommendation of the Tri-Gen report is that all production fish (i.e., non-broodstock) be 100% female and triploid (i.e., reproductively incapacitated). This recommendation was made: a) to prevent breeding, thereby addressing concerns about interbreeding with wild stocks; b) to protect property rights in the event a Newfoundland selection program resulted in a superior farming strain; and c) to prevent development of secondary sex characteristics in order to assure an aesthetically pleasing product.

The Dartmouth workshop on methods for the production of non-maturing salmonids was the beginning of a Science Branch initiative to evaluate this technology for potential application in Newfoundland. Scientists and producers from the international community made presentations to the workshop and conveyed the message that this technology has considerable potential to achieve the recommendations identified in the Tri-Gen report. One of the presentations to the Dartmouth workshop (Boulanger 1991) included experience with economic performance of triploid salmonids. The information presented indicated there is considerable economic gain to be obtained from triploid relative to diploid salmonids. However, one of the presentations (Benfey 1991) cautioned that the performance of triploid salmonids may be inferior in marginal environments. Two further presentations (Jungalwalla 1991; Sutterlin and Collier 1991) identified deformities among triploid stocks that may be a result of the triploid condition. There is some recent evidence that suggests triploid salmon may require dietary supplements of vitamin C.

As a result of the Dartmouth workshop, it appears that triploidy technology may have considerable potential for application in Newfoundland; demonstration of this technology under Newfoundland salmon farming conditions is essential. Salmonid farmers require objective criteria that will support business planning towards maximizing potential for profitability of Newfoundland salmonid farms.

It is in this context that Science Branch convened the present meeting to determine whether there is sufficient interest among the salmonid farmers to pursue a technology demonstration/transfer program. Such a demonstration would allow the Newfoundland farmers to make informed business decisions as to the potential benefits that they might expect in their own operations by making use of non-reproductive salmonids. From a Science Branch perspective, the goal of the present meeting is to develop an action plan for technology demonstration for which funding opportunities can be pursued in an attempt to implement a program this year.

With this as background, the chairperson opened the meeting to comments from the participants. A lengthy discussion followed (Appendix 3).

INDUSTRY REPRESENTATION

The salmonid farming industry was represented by businesses from five locations in Newfoundland (Fig. 1). Businesses represented were as follows:

Company	Location	Representative
Bay d'Espoir Salmon Growers Cooperative	St. Albans	Clyde Collier
Great Northern Peninsula Development Association	Daniel's Harbour	Gary Wilton
St. Mary's Bay Centre Development Association	Holyrood Pond	Sylvester Yetman
NewTech Char Farm	Port Rexton	Robert Freeman
Valley Char Ltd.	Grand Lake	Todd Turner

Fig. 1. Location map of Newfoundland salmonid aquaculture businesses

RESULTS

HIGHLIGHTS OF PARTICIPANT DISCUSSIONS

Industry Status

The salmonid farming industry in Newfoundland is using triploid rainbow trout and, in the context of continued involvement with this species, Bay d'Espoir salmon growers consider salmon to be their next priority. If present experiments with Atlantic salmon work out, and are economically feasible, demand for this species by the growers likely will escalate rapidly. There is also a requirement for more research on Atlantic salmon rearing techniques in the Roti Bay marine cages. Bay d'Espoir growers now place higher priority on evaluations of triploid salmon performance.

Sex reversal was undertaken at the Bay d'Espoir Hatchery early this year. As a result, juvenile inverted salmon, rainbow trout and Arctic char are available at the hatchery. The water temperature in the hatchery was 14°C all winter and the sex reversed groups grew rapidly. It is likely there will be one year smolts from these treatments in 1992 and that some of these may be about one pound in weight. Rates of growth are such that it is likely there will be some precocious maturation this fall. If so, there would be an opportunity to select sex-reversed females and use the homogametic milt to fertilize eggs from normal diploid females. If a pressure vessel is available with which to induce triploidy, there could be triploid parr in the hatchery next year.

Regardless of who the producers are or what species they are producing, their purpose is be economically viable. In the context of triploid Arctic char or brook trout aquaculture, if certified eggs are available, Newfoundland growers would be interested in evaluating them. However, if either the eggs or sperm are not certified, there is an element of risk to the producer. Newfoundland farmers have to build a business on what is available. The hatchery owners question if they can obtain the required all-female milt without risk to their present operations? Hatchery owners do not want to risk being decertified.

Industry Concerns

A concern was raised that, if the growers are dependent on reproductive sterilization for industry viability in Newfoundland, by the time they have adapted and perfected the technology under Newfoundland conditions, there will be no industry left in Newfoundland. The farmers are still interested in continuing with limited work on Atlantic salmon and are pursuing a cautious approach. This may change drastically as the market situation for salmon improves. Prices for Bay of Fundy salmon are improving and are now up to $4.30 per pound. If the shareholders in the Bay d'Espoir Salmon Growers Co-op find they can improve their profits by perhaps 20% relative to rainbow trout, the present 100,000 egg limit imposed by DFO will not suffice. In discussing reproductive sterilization, it is important to consider how it is going to support the industry. In supporting industry, one must determine what their priority species are. There is little choice but to pursue reproductive sterilization and what it may have to offer the industry. The species with the greatest potential for the Newfoundland industry at present are Atlantic salmon, rainbow trout and Arctic char. In considering further work with these species, one must not lose sight of the fact that there is a significant cost involved for the expansion of facilities and the provision of new apparatus. Since adequate triploidy technology has not yet been developed for Arctic char, there may be a significant delay in commercializing triploid production of this species. Technology, equipment and people are required to make this work. What is needed from this present forum is a frank and open discussion of perceptions and concerns out of which a "shopping list" of essential elements for a technology transfer/demonstration program for Newfoundland can be developed.

A further concern was expressed that triploid salmon may suffer undesirable side effects. In Tasmania, the main salmon producer is working with two triploid year classes, both of which have developed jaw deformities that are evident in about 20% of each year class. This has been attributed to the triploid condition. The deformities are not affecting feeding or growth rates. The Tasmanians suspect the problem is due to a vitamin C deficiency. They have found correspondence of jaw abnormality, triploidy and low vitamin C in the kidney.

The sea temperature in Tasmania reaches approximately 18°C during the summer. The fish grow rapidly but may be under some stress at these higher temperatures and the standard diet may not be adequate. These symptoms have not been found in diploid salmon. Deformities in these triploid fish have necessitated the carcasses being marketed as fillets. This is not desirable, considering that the best markets are for gutted, head-on salmon.

Considerable dissatisfaction was expressed by some of the participants who felt that DFO is not an advocate of aquaculture development. An objection was raised that, while there may be advantages to implementing a Newfoundland program with this technology, there will be a definite cost. The perception is that the whole concept of genetic "pollution" is theoretical fantasy. Why not just change the regulations? According to an industry spokesperson, the whole approach of DFO is designed to put the Newfoundland industry out of business at an early stage.

According to one industry spokesman, there are potential benefits to using triploid salmon. In Scotland, the farmers say they could not operate without this technology. However, the spectre of vague environmental impacts from genetic contamination is laughable in comparison to what gets past environmental regulations now. Science Branch has only a scientific possibility. The whole DFO business is madness! A representative of one funding agency indicated that there is criticism from managers of public funds that agencies should not consider support for activities that do nothing but address policy considerations that are unfounded in the first place. If DFO wants this technology, then DFO should pay for it.

The DFO response to this criticism was that regulations are there for a purpose. Ultimately, regulations may be changed but amendments would come about on the basis of knowledge, not pressure. In the absence of sufficient facts in this matter, the DFO approach has been to look for an avenue that has the greatest likelihood of success. Triploid technology offers a potential solution to everyone's needs. When the facts of the technology are known to the local farmers, if there is a significant benefit in terms of fish performance, the farmers will take advantage of it. If not, they will not use it. The present effort is simply to provide the basis for that decision on their part.

Essential Elements of a Newfoundland Program

A consideration that must be addressed prior to determining the scope of a Newfoundland program on non-maturing salmonids is the source of gametes to support the program. Importation into Newfoundland is highly dependent on quarantine. Currently, any eggs shipped to Newfoundland must be from disease free parents and must go directly into an approved quarantine facility for hatching. Juvenile fish must remain in quarantine for a period of four months after first feeding. Disease analysis of samples of these juveniles must indicate that the group can be certified under the FHPR Schedule II before the fish can be removed from quarantine.

DFO is governed by these sorts of constraints in trying to identify the means whereby a technology demonstration program could be implemented. While this complicates the planning process, there still are several constraints that would have to be met. However, as a first step in addressing the constraints, the following sequence of events is consistent with the intent of the Fish Health Protection Regulations (FHPR) and could be undertaken under Section 4 of the Fishery Act:

Procurement of unfertilized salmonid eggs: All eggs must be from an approved site. Female salmon are to be stripped into sterile containers and each female salmon used must be sacrificed for complete analysis under the FHPR protocols. All lots of eggs are to be labelled to identify the female parent that produced the particular lot of eggs.

Shipping of unfertilized salmonid eggs: Eggs are to be transferred directly into an approved quarantine facility. Once in the quarantine facility, eggs are to be removed from the transfer container and the container, as well as all fluids and miscellaneous shipping materials, are to be chlorinated for a minimum

of 24 hours.

Sperm collection: Salmonid sperm will be stripped into sterile containers outside of the quarantine facility. Only the sperm and the containers are to enter the quarantine facility. All sperm donors will be sacrificed for complete FHPR analysis. Sperm containers will be labelled with the identification number of the sperm donor. Care must be taken whenever the gonad is being surgically removed (i.e., for sex-reversed milt) that surgical equipment is sterilized before proceeding to another specimen.

Fertilization of salmonid eggs at the recipient quarantine facility: Fertilization is to take place in the quarantine facility in a recirculation system. Any wastes are to be chlorinated. All water-hardened zygotes are to be surface disinfected before incubation. Incubation is to take place in a recirculation system. All equipment used during fertilization (i.e., gamete containers, fertilization pans, water hardening apparatus) is to be chlorinated before removal from the quarantine facility.

Consequences of FHPR analysis: If FHPR pathogens are found in either of the parents for a particular mating, the resulting lot of zygotes is to be destroyed. Progeny from disease-free eggs are to be retained in the quarantine facility for four months subsequent to "first-feeding". Parr samples are to be subjected to FHPR analysis after four months of active feeding. If any bacterial or viral FHPR agents are found in parr, all are to be destroyed.

What is required, in a technology transfer and demonstration program, is a mechanism whereby the Newfoundland salmonid farmers will have access to first hand information that will allow them to come to their own conclusions about this technology and whether it offers them any advantages. This sequence may provide the beginnings of that mechanism.

A caution, expressed by the consulting community, was that industry must be careful regarding the species to be included in the program. Techniques are available for salmon and rainbow trout but procedures have not been worked out for Arctic char. There are means to get into a demonstration program with Atlantic salmon and rainbow trout. With char, research is needed to figure out reliable techniques to get effective triploidy. A further caution was presented by Pepper (1991a):

> "When the triploidy method is used, it is essential to use all-female stock in order to avoid growth and behaviour problems related to the sexual maturation of males. While the Y-specific, DNA probe technology being developed for Atlantic salmon by Dr. R. Devlin (DFO, CODE for Biotechnology & Genetics, West Vancouver Laboratory) likely would be the preferred means to assure XY males are removed from the broodstock, this technique may not be available in time. In the absence of this technology, the methods of Jungalwalla (this workshop) may have to be used. This procedure follows:
>
> Each presumptive donor (i.e., treated fish showing all-male secondary sexual characteristics) is squeezed, and accepted only if no milt is expressed. If upon dissection the testes appear normal, the fish is again rejected. Only if the testes are lobulated is the fish accepted. The presence of only one lobulated testis, and/or the presence of a vestigial ovary, increases confidence that the donor is indeed a masculinized female."

Research Opportunities

Salmonid farming industry development needs present three research opportunities that should be incorporated into a technology transfer/demonstration program: 1) salmonid maturation monitoring and refinement of maturation control technology; 2) genetic characterization of stocks; and, 3) microbiology and examination of disease agents.

Salmonid maturation: the primary orientation is to implement a program to evaluate the performance

of triploid salmonids relative to normal diploids. This provides the opportunity to monitor the physiological progression of the maturation process and to quantify how this progression in diploids differs from the experimental triploid group;

Genetics: the DFO has advocated that local Atlantic salmon stocks be used for brood development. The salmon farmers have been using St. John River stock from the Bay of Fundy sea cages. With this to draw on, factors in the experimental design for performance evaluations will be stock (i.e., local Newfoundland and St. John River) and ploidy status (i.e., 2n and 3n). Such a factorial design would present the opportunity to examine the biochemical genetics of at least four groups of salmon in the search for appropriate fragments of the genome with which DFO might be able to substantiate its concerns about maintaining genetic variability among wild salmon stocks.

Disease: the DFO concerns about salmon stocks (i.e, fish disease and genetics) are identified in the Newfoundland Fishery Regulations. The Canadian FHPR provide the protocols with which to screen salmonid populations for pathogens of concern. However, industry questions how much research DFO actually commits to microbial chemistry and etiology of the microorganisms identified in the FHPR. The present farming industry, as well as the required demonstration program for the initiative on non-reproductive salmonids, will provide DFO with a valuable opportunity to undertake additional research in each of these fields.

Infrastructure Requirements

If industry is to proceed, a pressure vessel will be needed to induce the triploid condition. Since these vessels operate under high pressure, industry should consider looking for the necessary expertise to operate the pressure vessel. To summarize the situation, industry now has: 1) a potential supply of homogametic milt at Bay d'Espoir; 2) a quarantine facility at Bay d'Espoir; 3) a certified hatchery at Port Rexton; and, 4) definition of the prerequisites to allow industry to bring in unfertilized eggs. What is needed is: 1) a pressure vessel; and, 2) additional tank space.

RECOMMENDATIONS

Recognizing that this is a government-industry initiative, and that the pressure vessel represents a common need for Newfoundland work with triploid salmon, the Federal and Provincial governments should collaborate on acquisition of a pressure vessel and the expertise to use it. On the other hand, it is not likely that government would propose specific facilities for individuals, who may or may not want to be involved in such a program. Therefore interested growers will develop their own proposals, for submission under the auspices of a Newfoundland demonstration program on non-maturing salmonids.

Having decided on the number of smolts required for comparative performance evaluation, one can calculate, if there is knowledge of what losses to expect from triploid eggs, the number of eggs required for import. The 100,000 egg limit was defined on the basis of genetic concerns. Therefore, in considering triploid eggs, or potentially haploid eggs that are to be imported, fertilized with milt of sex-reversed females, and pressure shocked, the requirements are likely to be incremental to the present 100,000 limit.

An important feature of the infrastructure proposals is the level of funding that will be required. However the proposals are developed, they should be linked and submitted in reference to the Dartmouth workshop proceedings document (Pepper 1991a). This would help provide substantial validity within the government circles that would be involved in reviewing the proposals. It is important to convey that the proposals are in line with the overall needs of a technology demonstration/transfer program for triploid salmonids.

For the moment, the planning initiative is concerned only with program implementation. The duration

of the program will depend on the results and the number of species that ultimately are included in a demonstration program. This may take several years since it is quite likely industry will want to make refinements to the program as it unfolds. The first step will be the proposal for the pressure vessel. This will be pursued between the two levels of government during July. The proposals for appropriate support facilities are to be submitted to Brian Meaney (Department of Fisheries, Government of Newfoundland and Labrador) or Vern Pepper (DFO, Government of Canada) by the week of August 26 - 30. In the interim, industry will be provided with the minutes of this meeting and subsequently, with the outcome of the various proposals.

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I would like to express my appreciation to the DFO Industry Development Division for allocating funding from the Development component of the Atlantic Fisheries Adjustment Program in support of this industry consultation forum. I also thank the participants for their time and their support for this initiative. Terry Nicholls (DFO, Science Branch) assisted in preparation of this document; Dr. Derek Shaw and Dave Scruton reviewed the manuscript and provided many helpful suggestions. Dr. Tillmann Benfey (University of New Brunswick) saved me from some potentially embarrassing deficiencies in the technical definitions of Appendix 2.

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APPENDIX 2. EXPLANATION OF TECHNICAL TERMS

all-female	A population comprised entirely of individuals that will develop ovaries during sexual differentiation and maturation.
chromosome	Rod shaped bodies contained within the cell nucleus. Chromosomes are made of DNA and at least two kinds of proteins. Genes are arranged in linear order on the chromosomes.
DNA	Deoxyribonucleic acid.
family-tie	A breeding scheme to maximize genetic ties between families so that the relationships between fish resulting from the breeding plan facilitate estimation of breeding value. Each male is mated to three females and each female contributes to either one or two families. A family-tie model using four males and eight females results in 12 families for which performance (i.e., growth, survival, food conversion) can be monitored for evaluation of breeding value of individual families relative to the other families.
genome	The minimum group or set of chromosomes contained within a zygote.
gamete	A mature sex cell that has half of the chromosomes found in each cell of an organism. Normal male sex cells (i.e., spermatozoa) have either a Y chromosome or an X chromosome. Female sex cells do not have a Y chromosome. When two gametes fuse (i.e., an egg and a spermatozoon), the resulting zygote will be either XX (female) or XY (male) according to whether the sperm had an X or a Y chromosome.
gene	A unit of heredity. Composed of DNA and located on a chromosome.
homogametic	Referring to an organism, or the gametes of an organism, having only one type of sex chromosome.
inverted	Organisms, or the gametes of an organism, that have been manipulated to yield the opposite sex from that predicted by normal development. In several species of fish, this is accomplished by a process that requires two generations. In the first generation, young fish are immersed in or fed male sex steroids. This confuses the normal gonad development so that females develop male gonads. These fish are genetic females (i.e., have XX sex chromosomes) but resemble males. All the normal males (i.e., X and Y sex chromosomes) are eliminated before reproduction. Sperm is then collected from the manipulated females and used to fertilized the eggs of normal females (i.e., all X) so that the resulting zygotes are all females. Since female salmon and trout typically do not mature as early as males, all-female salmonids have better performance under aquaculture conditions.
milt	Fish sperm.
ploidy	The number of complete chromosome sets in a cell nucleus. Most organisms have two sets of chromosomes (i.e., 2n), one set from each of the two parents.

haploid - 1n. One set of chromosomes. Typical of gametes. Fusion of egg and sperm results in a 2n organism.

diploid - 2n. Two sets of chromosomes. Typical of most organisms.

triploid - 3n. Three sets of chromosomes. This situation is unusual and, among salmonids, results in a reproductively sterile organism due to the fact that the reduction division that takes place to form a haploid gamete becomes incapacitated. In salmonid aquaculture, maturation of diploid fish results in diversion of food energy into gonad development rather than growth. In order to assure that food energy is utilized only for growth, salmonids are reproductively incapacitated by manipulation to a 3n chromosome complement. This often is accomplished by subjecting newly fertilized, all-female eggs to high pressure.

pathogen A microorganism that may cause disease.

plasma The liquid, intercellular component of blood.

precocious maturation
Production of viable gametes prior to attaining normal adult characteristics.

quarantine In the context of fish health, this is a facility that is free of pathogens (i.e., no pathogens in the incoming water and no pathogens in the waste water).

seminal Pertaining to or associated with an expanded portion of the genital duct of the male; functions for storage of spermatozoa or secretion of accessory substances.

sex reversal The process of changing the sex of an organism (see "all-female" and "inverted").

spermatozoon A male sex cell (plural is spermatozoa). The mixture of spermatozoa and seminal plasma is called sperm.

strain A group of organisms of the same species that has genetic characteristics that are particular to that group and which differ from other groups of the same species.

tannins Organic chemicals found in many plants. These substances are soluble in water and often are released from decomposing vegetation. These are common chemical components of wetlands and often impart a dark brown colour to waters that drain from bogs. These chemicals are used in tanning animal skins.

zygote A fertilized egg before the beginning of cell division.

APPENDIX 3. PARTICIPANT DISCUSSIONS

At the request of the chairperson, C. Collier and A. Sutterlin provided an update on work that has taken place at Bay d'Espoir and their efforts to date relative to triploid salmonids.

Sutterlin Sex reversal was undertaken at the Bay d'Espoir Hatchery early this year. As a result, we now have inverted salmon, rainbow trout and Arctic char. The water temperature in the hatchery was 14°C all winter and the sex reversed groups are growing rapidly. It is likely there will be one year smolts from these treatments in 1992 and that some of these may be about one pound in weight. They have been growing fast enough that it is likely there will be some precocious maturation this fall. If so, there would be the opportunity to select the sex-reversed females and use homogametic milt to fertilize eggs of normal diploid females. If a pressure vessel is available with which to induce triploidy, there could be triploid parr in the hatchery next year.

Collier The Bay d'Espoir growers have been involved with Atlantic salmon, rainbow trout, and Arctic char. Rainbow trout is the preferred species simply because it causes the least amount of grief in our cage operations. A condition of our import license from DFO was that the rainbow eggs be all-female, triploid. The growers are most anxious for us to develop our own rainbow trout brood stock. With no brood stock of our own, we are dependent on a mainland supplier. As it is, if the present supplier runs into problems, our rainbow trout farmers will be out of business. As well, we have seen ~7% mortality among sac fry due to failure to resorb the yolk sac. Added to this, we have seen some spinal deformities. While we realize these problems may be due to the triploid condition, we fear that it could be due to limited numbers of brood and corresponding inbreeding. We consider our future with rainbow trout is dependent on developing our own brood even though we expect to continue importing eggs for the next few years.

We have experienced several quirks with Atlantic salmon that are yet to be overcome. At present, we are concerned about extremely dark body coloration of salmon from our marine cages. This seems to be caused by the abundance of tannins in Roti Bay water. We are trying to evaluate this situation through our marine cage experiment in Gaultois Passage (Strickland Cove). In the mean time, while we are relying on our rainbow trout to keep us going, we continue to be interested in Atlantic salmon simply because, on paper, the economics of salmon are much better than for trout. For the present membership of the Salmon Growers Cooperative, the 100,000 egg importation limit for St. John River stock, that was imposed by DFO, is sufficient to meet our immediate needs. However, there may yet be a request from a business interest in Gaultois, as well as from the Conne River Native Band Council, for Atlantic salmon. If this transpires, we expect our need for salmon eggs will greatly exceed the 100,000 limit. For the time being, we wish to proceed cautiously with Atlantic salmon experiments in an attempt to resolve our present culture quirks and therefore avail of the better economic outlook for salmon relative to rainbow trout.

Brown What other restrictions are there on St. John River fish? Can you not develop a brood stock from the salmon already imported?

Pepper Eggs of the St. John River stock were imported with the following conditions: 1) the eggs and resulting fry be confined to the hatchery until smoltification; 2) a transfer license be obtained prior to moving smolts into marine cages; and, 3) the salmon resulting from the egg importations not be used for brood stock.

Brown Is this a DFO restriction or a restriction imposed by the egg supplier?

Pepper DFO imposed these conditions.

Collier The salmon farmers would prefer to obtain eggs from their own stock. At $0.15 per egg, importation is costing Newfoundland salmon farmers a lot of money.

Brown In order to implement a program on non-maturing salmonids, would these DFO importation conditions have to be lifted?

Pepper If a comparative performance evaluation is required to evaluate triploid salmon relative to diploid salmon, the experiments may not require a lot of fish. The present experiments with Grand Codroy relative to St. John River stock are based on about 9,500 smolts of each stock. This required an initial egg supply of ~20,000 per stock. If eggs are subjected to pressure shock to induce triploidy, additional egg mortality of perhaps 30% could be expected. Even if 50,000 eggs were required to produce some 10,000 smolts for performance evaluation, this still fits within the 100,000 import limitation.

Collier We want also to look at the economics of raising these fish and therefore require sufficient smolts to do an adequate evaluation of the economics of raising the treated fish.

Penney The limit of 100,000 eggs was imposed in consideration of potential genetic impacts on wild populations. As long as eggs are free of certifiable diseases, and the resulting fish are not able to interbreed with local wild stocks (i.e., they are triploid), the 100,000 limit should not apply. Therefore, it is not a question of lifting the import limitation since this limit was defined on the basis of reproductive concerns.

Brown DFO has legislation and policy concerns relative to fish health and genetics. Does salmonid enhancement fall within the same policy and legisiative concerns?

Pepper There are no exclusions for salmon enhancement.

Brown We know there are changes in stock gene pool in enhancement situations. Stripping, incubation, and hatchery rearing change selection pressures and therefore, inevitably result in genetic shifts within the populations with which you are working.

Pepper We are very aware of the dangers inherent in our enhancement projects. Work by Dr. Ruth Withler at our Pacific Biological Station has demonstrated the need for procedures to maximize stock genetic variability. We know our stock breeding goals in enhancement are very different from breeding goals in aquaculture. For aquaculture, we need a "big, dumb fish" that likes people. For enhancement, we need an agile, wiry, wary fish that avoids anything bigger than itself. Knowing that we need to maintain genetic variability in our enhancement operations, we go to considerable effort to maintain a satisfactory "effective population size" and to assure cross fertilization among parents of our enhancement brood stocks. In Newfoundland, where we rely largely on distribution of swim-up fry to natural habitats, we avoid the concerns of hatchery conditioning during the freshwater residence interval. While we do not discount the possibility of inadvertent erosion of genetic variability in our enhancement operations, we do take steps to minimize this possibility.

Moir What has been the response from Scotland on the performance of triploid salmon?

Pepper The latest information I have from Scotland is that triploid salmon are performing well and the farmers are obtaining significant economic gain from triploid stock relative to reproductively active stock.

Sutterlin In Tasmania, the main salmon producer is working with two triploid year classes, both of which have

developed abnormalities that are evident in about 20% of each year class. This has been attributed to the triploid condition. The deformities are not affecting feeding or growth rates. The Tasmanians suspect the problem is due to a vitamin C deficiency. They have found correspondence of jaw abnormality, triploidy and low vitamin C in the kidney. The sea temperature in Tasmania runs about 18°C during their summer. Their fish are growing very rapidly and may be under some stress at these temperatures. Under these conditions, the standard diet may not be adequate. These symptoms have not been found in diploid salmon. Deformities in these fish have necessitated that the carcasses be marketed as fillets.

Collier It is very difficult to sell fillets in the U.S. The Americans want to do the processing themselves. Fillets could be sold in Canada but the best market price is in the states. However, there may yet be an advantage, even to triploid fish that have to be processed as fillets. The St. John River salmon have a 10-12% grilsing rate. If grilsing is eliminated through triploidy, we may be able to offset the "cost" of the deformity problem.

Pepper The Bay d'Espoir Salmon Growers Cooperative has taken the necessary steps to implement experiments with triploid salmon, rainbow trout and Arctic char. Therefore, it is likely work can continue there this fall. Are there other growers who are interested in participating in performance evaluation experiments who can be ready to start this fall?

Freeman While I am not interested in Atlantic salmon, I would like to be involved with Arctic char and brook trout.

Yetman We are very interested in becoming involved in a program with Atlantic salmon at the land-based facility at Holyrood Pond. Unfortunately, satisfactory smolts have never been available to us. The Holyrood Pond facility is not operational at the present time.

Pepper A consideration that must be addressed prior to determining the scope of a Newfoundland program on non-maturing salmonids is the source of gametes to support the program. Importation into Newfoundland is highly dependent on quarantine. Currently, any eggs shipped to Newfoundland must be from disease free parents and must go directly into an approved quarantine facility for hatching. Juvenile fish must remain in quarantine for a period of four months after first feeding. Disease analysis of samples of these juveniles must indicate that the group is free of certifiable diseases before the fish can be removed from quarantine. Also, the Newfoundland Region, Introductions and Transfers Committee, in consideration of the pathogen history of the Bay d'Espoir fish culture facilities, is not likely to support transfer of smolts to the Holyrood Pond site from Bay d'Espoir (Department of Fisheries and Oceans. 1990).

Sutterlin Does this mean that the Bay d'Espoir Hatchery would have to be certified to provide smolts to the rest of the province.

Pepper In essence, virtually; in fact, no. At present, Bay d'Espoir Hatchery smolts can be transferred only to marine farms within the Bay d'Espoir estuary because of the known presence of pathogens in the Bay d'Espoir area.

Meaney Hatchery certification, as defined in the FHPR, pertains only to interprovincial transfers. The FHPR, and therefore the "certification" status does not apply for intraprovincial transfers.

Pepper True. The term "certified" is a poor choice of words. However, in the present context, the notion is correct though perhaps it is too rigorous. Since we know there are pathogen concerns at Bay d'Espoir, the "spirit" of the Newfoundland Fisheries Regulations is to prevent spread of these

pathogens.

Meaney There is no mandatory exclusion of intraprovincial transfer of smolts from the Bay d'Espoir Hatchery. If it can be shown that the same diseases are present at the proposed recipient site, there is no reason or justification for prohibiting the transfer.

Pepper My concern with disease is that I would not want to encourage people to become involved in a program with non-maturing salmonids if that involvement could jeopardize the fish health status of their facility.

Yetman We have been told by Kenebec Aquaculture of Maine that they will be able to provide us with 1000-1500 salmon smolts in 1992 that will meet all Canadian import regulations. Since the Holyrood Pond facility is land-based, there is little chance of escape to the wild and therefore, no concern about genetics. Regardless, we may be interested in participating in a demonstration program with non-maturing Atlantic salmon at that time.

Pepper Your land based facility may provide access to certified stock from outside Newfoundland. However, involvement in a program with triploid salmon still requires consideration of the logistics of how that involvement could come about. If triploid eggs are available from a disease free facility, there may not be a problem getting started. However, if all-female, triploid eggs are not available from a certified facility, you still would require access to a quarantine facility into which to import haploid eggs and homogametic milt. I do not have any immediate answers and would have to check on the prerequisites with our fish health people.

Brown In attempting to resolve the conflicts between your DFO mandate problems and salmonid farming industry development needs, you should bear in mind that industry needs present you with three research opportunities: 1) salmonid maturation monitoring and refinement of maturation control technology; 2) genetic characterization of stocks; and, 3) microbiology and examination of disease agents.

Salmonid maturation: Of course, your orientation here is to implement a program to evaluate the performance of triploid salmonids relative to normal diploids. This provides you with the opportunity to monitor the physiological progression of the maturation process and to quantify how this progression in diploids differs from your experimental triploid group;

Genetics: DFO has been anxious to have local Atlantic salmon stocks used for brood development. The salmon farmers have been using St. John River stock from the Bay of Fundy sea cages. I assume therefore that, ultimately, factors in your experimental design for performance evaluations will be stock (i.e., local Newfoundland and St. John River) and ploidy status (i.e., 2n and 3n). Such a factorial design would present you with the opportunity to examine the biochemical genetics of at least four groups of salmon in the search for appropriate fragments of the genome with which DFO might be able to substantiate its concerns about maintaining genetic variability among wild salmon stocks.

Disease: As you pointed out in your opening presentation, DFO concerns about salmon stocks (i.e, fish disease and genetics) are identified in the Newfoundland Fishery Regulations. Your Canadian FHPR provide you with the protocols with which to screen salmonid populations for pathogens of concern. However, I question how much research DFO actually commits to the microbial chemistry and etiology of the microorganisms identified in the FHPR. I suggest the present farming industry, as well as the demonstration program you are advocating, will provide DFO with a valuable opportunity to undertake additional research in each of these fields.

Pepper Your points are well taken. I agree that the factorial experiment you advocate for a demonstration program on non-maturing salmonids would satisfy our need for objective data on local vs. imported stocks, and triploid vs. diploid performance under Newfoundland salmonid farming conditions. However, I rather expect we will not have access to the means to implement a program at the level you suggest within the immediate future. If we are to implement a program this year, we will have to confine ourselves to the St. John River stock since the only homogametic milt we may have access to this fall will be from St. John River precocious parr. We have an experiment underway now with Grand Codroy stock but these are normal diploids. We would not have access to homogametic milt from the present experiments until 1994 at the earliest. Therefore, I expect we will have to confine ourselves to the St. John River stock for the next few years. While this may not orient us towards a comprehensive breeding program to develop the best stock in the shortest time, the smaller experimental design will provide an industry perspective on how triploids perform relative to diploids.

I find your advice on maturation monitoring and genetics research to be particularly rewarding. This work is being done even with the present experiments in Roti Bay on performance evaluation of Grand Codroy vs. St. John River stocks. These experiments started in May of this year. Starting in March of next year, I will undertake a blood sampling program to monitor the onset of maturation and to provide the material for further work on biochemical genetics. Dr. Idler, of the Ocean Sciences Centre, has agreed to do vitellogenin assays of blood serum samples and Dr. Davidson (MUN Biochemistry Department) will take the red blood cells for DNA fingerprinting. In some of our previous work with Dr. Davidson, through the Science Subvention Program, Dr. Davidson has recommended,

"The time has come for salmon population geneticists to turn their attention to the identification and isolation of the genes that give rise to selectable traits (e.g. genes for age at sexual maturity; age of smolting; disease resistance; pH tolerance, etc.)."

While implementation of a structured brood stock breeding program would provide the most appropriate vehicle with which to pursue Dr. Davidson's recommendation, you are correct in suggesting that the proposed program offers considerable opportunity for this sort of research. These opportunities are being pursued.

Finally, I confess that I had not considered the proposed experiments as an opportunity for work on fish health. Again, this is a constructive suggestion that I will pursue during the next several months. I am particularly intrigued by the prospect of looking at stock adaptation to diseases. Is it likely that we will see that local stocks have greater resistance to local diseases? If so, the proposed experiments will provide a fine opportunity to look at the possibility of different strains of infectious microorganisms and at immune system response to these disease agents.

Moir I understand the FHPR are under review. Therefore, how much of our discussions are likely to be redundant under the revised regulations?

Pepper I doubt that very much of what we have discussed will be redundant in that the FHPR were developed with the intent of preventing the spread of diseases that are harmful to salmonids. This intent is not likely to change. I anticipate extensive revisions in the form of refinements that will achieve the intent in as fair and equatable a manner as possible. If we define our demonstration program within a framework that is responsible relative to confining and curtailing the spread of salmonid diseases, then it is quite likely our plan will fit within any amendments to fish health regulations.

Penney Does the quarantine facility at Bay d'Espoir sterilize its incoming water?

Collier No.

Sutterlin It would be relatively simple to sterilize incoming water through the incubation interval. Beyond first feeding, the hatchery just does not have the capability.

Penney That is why fish cannot move outside of the Bay d'Espoir area. We know there are diseases of concern in the hatchery water supply and therefore know the fry in the hatchery and quarantine facility may be exposed to these diseases. Since we know there are pathogens of concern in the Bay d'Espoir estuary as well, the whole system, from incubation to marine cages, is a potential concern.

Turner Do the same regulations apply for eggs from wild stocks?

Meaney For a wild stock, fertilized eggs are held in quarantine. All parents are killed for disease analysis. If the parents are clean, the eggs are automatically certified.

Sutterlin What is likely to be DFO's attitude towards stocking of public waters with sterile rainbow trout?

Pepper Even having wondered about this myself, I still must say that I do not know. However, proceeding from biological principles, since there should not be long-term genetic impacts, concerns likely would focus on more immediate ecological impacts of competition and predation. In the absence of definitive studies of behavioral interactions, I anticipate a cautious approach. I would expect some sort of experimental approach, perhaps using a pilot project in which potential impacts could be monitored.

Meaney There are several proposals being considered by Provincial Fisheries right now. There are about 15 people proposing fish-out ponds for rainbow trout as part of tourism development. Many of these proposals are in conflict with the published DFO "Policy for Introductions and Transfers of Salmonids in the Province of Newfoundland and Labrador". While some of these proposals are only at the stage of posing the questions, some proponents have invested considerable amounts of money evaluating water supplies and project logistics. In cases where these proposals are for natural water bodies, there is likely to be conflict with DFO.

Pepper An artificial pond is not likely to elicit the same level of concern as a natural pond provided there are adequate precautions to prevent the fish from escaping from the artificial situation. Part of the difficulty here is that people are looking for categorical statements from DFO. We have very few absolutes when it comes to biology. While we can proceed from a general and common ground such as the concepts behind the policy for introductions and transfers, the specifics have to be considered on a case by case basis. We have to look carefully at the notion of risk assessment. High risk warrants greater caution. Added to this is the difficulty of quantifying risk which, most often, is not well defined. There has been some progress towards a "vernacular science" but this is a relatively new area of endeavour and likely will require considerable refinement if it is to be an adequate tool for risk assessment.

Meaney It seems to me there are three main reasons why we all should be involved in this technology for reproductive control:

1) Meet government regulations.

The easiest way to get around the regulatory problems you have referred to is to change the regulations and therefore not have to spend any money. As you indicated, the province also has legislation governing importation of stock and this legislation could require environmental assessment under the Environmental Assessment Act. Licenses have been issued to import St.

John River stock to Bay d'Espoir because no provincial department has ever identified an environmental concern.

2) Prevent Environmental Impacts.

Are we looking at reproductive sterilization to prevent environmental damage? Everybody talks about the threat and potential consequences of genetic "pollution". It would be worth while if someone took the time to document what is known on this issue. I attended a workshop in Norway on this topic last year. It was apparent there was considerable disagreement within the scientific community as to the consequences of genetic intermixing. While it likely is politically correct to assume there could be some environmental concern, such an assumption has a pronounced negative impact on industry development. It is the farmers who are paying for this concern. They have the most to lose.

3) Economic Return.

A more positive approach is to look at the potential for improved economic return from a non-maturing salmonid. An example is from experience with brook trout in Quebec where the farmed product was not marketable due to maturation. The maturing trout looked horrible and the flesh was inferior and unacceptable to the consumer. In this context, there is likely to be a whole lot of interest within industry for further work with this technology as well as participation in an evaluation program. Any approach that conveys an advantage to the growers is likely to be embraced by industry.

My concern is that, if we are dependent on reproductive sterilization for industry viability in Newfoundland, by the time we have adapted and perfected the technology under Newfoundland conditions, there will be no industry left in Newfoundland. We have heard that the farmers are still interested in continuing with limited work on Atlantic salmon and are pursuing a cautious approach. We can expect this to change drastically as the market situation for salmon improves. Prices for Bay of Fundy salmon are improving and now are up to $4.30 per pound. If the shareholders in the Bay d'Espoir Salmon Growers Co-op find they can improve their profits by say 20% relative to rainbow trout, the present 100,000 egg limit imposed by DFO will not suffice. In talking about reproductive sterilization, we have to talk about how it is going to support the industry. In supporting industry, we have to determine what species have priority for industry. We have very little choice but to pursue reproductive sterilization and what it may have to offer the industry. The species with the greatest potential for the Newfoundland industry right now are Atlantic salmon, rainbow trout and Arctic char. In considering further work with these species, we must not lose sight of the fact that there is a significant cost involved for expansion of facilities and the provision of new apparatus. Technology, equipment and people are required to make this work. What we need now in this present forum is a frank and open discussion of perceptions and concerns out of which the chairman can develop a "shopping list" of essential elements for a technology transfer/demonstration program for Newfoundland.

Pepper What I am looking for in a technology transfer and demonstration program is a mechanism whereby the Newfoundland salmonid farmers will have access to first hand information that will allow them to come to their own conclusions about this technology and whether it offers them any business advantages.

Moir There may be advantages to implementing a Newfoundland program with this technology. There will be a definite cost. Why bother? The whole concept of genetic "pollution" is pretty wishy-washy. Why not just change the regulations?

Pepper The regulations are there for a purpose. Ultimately, they may be changed but it would come about

on the basis of knowledge, not pressure. In the absence of sufficient facts in this matter, my approach has been to look for an avenue that has the greatest likelihood of success. I see this technology as offering a potential solution to everyone's needs. When the facts of the technology are known to the local farmers, if there is a significant benefit in terms of fish performance, the farmers will take advantage of it. If not, they will not use it. The present effort is simply to provide the basis for that decision on their part.

Moir — The whole approach of DFO is designed to put us out of the game at an early stage. There are potentially some benefits to using triploid salmon. In Scotland, the farmers say they could not operate without triploid salmon. They receive a real financial benefit. This is positive. However, do not talk to us about environmental impacts. It is laughable when you consider these vague, airy-fairy sorts of genetic impacts, relative to what gets past environmental regulations now. There are advantages to triploidy and you are putting yourself at a distinct disadvantage by indicating you are concerned about regulations. Don't talk about concerns. Don't talk about regulations. You are advocating expenditure of considerable amounts of money to address something that may not even be a problem. Meanwhile, we have an industry that is struggling to get ahead. Your concern is imagined. You have a scientific possibility. The industry is real. It has real problems that must be addressed. The whole DFO business is madness!

Meaney — No one in DFO has come out and identified the rationale of why we should follow this route. Something is needed from the Newfoundland Region to back up regional policy statements on introductions and transfers.

Pepper — I believe what you are looking for is covered in my Dartmouth workshop presentation entitled "Motives, Actions and Goals...". The references quoted in my workshop presentation, especially the NASCO papers, provide some of the basis for DFO concerns.

Brown — All of the evidence to date is very inconclusive and therefore your policy is on very shaky ground. If you try to sell the policy on a shaky base, your delivery also will be shaky.

Moir — I face the criticism from my managers that we should not consider funding activities that do nothing but address policy considerations that are unfounded in the first place. If DFO wants it, then DFO should pay for it.

Collier — Why is Newfoundland Region making such a big thing of all of this when other regions are not?

Pepper — There are many regional differences across Canada; different species, different environments, different community complexities. We have a Fisheries Act to provide the principles of DFO mandate. We also have regulations made pursuant to the Act that allow for explicit recognition of regional dichotomies. The Newfoundland Fishery Regulations provide the basis for departmental concerns in this province. As well, Newfoundland Region has been concerned about consequences of importations to Newfoundland because this is where the impacts would take place. If mainland salmonid farmers wished to import Newfoundland stocks, the concerns would arise on the recipient end.

Halfyard — It appears though that regulations are being interpreted in different ways among the different regions.

Moores — We need to be aware that there are regulations, that tend to be of a relatively general nature, and there are policies that are the means whereby the regulations are administered in the various regions. All regulations are subject to ongoing amendments and amendments are common. If the aquaculture

industry can show that a regulation is unfounded, then the regulation can be amended. One has to recognize that regulations are there for a purpose and if the regulation is not well founded, it can be changed. A policy is much easier to change than a regulation but the door is never closed in terms of regulations.

Pepper I would like you to be aware that my normal sphere of activity within DFO is in Science Branch. Therefore, I have very little input into writing or amending regulations. Neither is it my function to motivate development since that role falls within the DFO Industry Development Division. While I occasionally have the opportunity to interact with people within the wider DFO mandate, such as this meeting under the Development Component of AFAP, my immediate responsibility is to Science and to issues for which Science advice is required. Our efforts to date with Dr. Davidson of the MUN Biochemistry Department have not resolved the genetic issue and therefore, I have no objective base for recommendations on this issue. However, as a result of the Dartmouth workshop, that was undertaken by Science Branch and funded under the Science Component of AFAP, I do have the opportunity to recommend pursuit of a Newfoundland program to demonstrate reproductive suppression technology under Newfoundland salmonid farming conditions.

Freeman You have talked mainly about Atlantic salmon and rainbow trout. I would be more interested in Arctic char and brook trout. Are you considering all species?

Pepper Yes but in a relative sense. The following hierarchy of priority was stated in the Dartmouth workshop proceedings document: 1) Atlantic salmon; 2) rainbow trout; 3) Arctic char; and, 4) brook trout. I suspect our efforts with these various species will be determined by the funding resources available to us.

Penney There is considerable interest in Newfoundland in fish-out pond development, particularly for brook trout. I have been in contact with a brook trout hatchery in Quebec that may have triploid brook trout eggs available this fall for limited research. If they do become available, we may look for some funding to get started with this species.

Sutterlin How could we proceed if we wanted to import unfertilized eggs into Newfoundland for fertilization with local, homogametic milt?

Pepper There are several constraints that would have to be met. However, I understand the following sequence of events is consistent with the intent of the FHPR and could be undertaken under Section 4 of the Fishery Act:

Procurement of unfertilized salmonid eggs.

All eggs must be from an approved site. Female salmon are to be stripped into sterile containers and each female salmon used must be sacrificed for complete FHPR analysis. All lots of eggs are to be labelled to identify the female parent that produced the particular lot of eggs.

Shipping of unfertilized salmonid eggs.

Eggs are to be transferred directly into an approved quarantine facility. Once in the quarantine facility, eggs are to be removed from the transfer container and the container, as well as all fluids and miscellaneous shipping materials, are to be chlorinated for a minimum of 24 hours.

Sperm collection.

Salmonid sperm will be stripped into sterile containers outside of the quarantine facility. Only the sperm and the containers are to enter a quarantine facility. All sperm donors will be

sacrificed for complete FHPR analysis. Sperm containers will be labelled with the identification number of the sperm donor. Care must be taken whenever the gonad is being surgically removed (i.e., for sex-reversed XX milt) that surgical equipment is sterilized before proceeding to another specimen.

Fertilization of salmon eggs at the recipient quarantine facility.

Fertilization is to take place in a quarantine facility in a recirculation system. Any wastes are to be chlorinated. All water-hardened zygotes are to be surface disinfected before incubation. Incubation is to take place in a recirculation system. All equipment used during fertilisation (i.e., gamete containers, fertilization pans, water hardening apparatus) is to be chlorinated before removal from the quarantine facility.

Consequences of FHPR analysis

If FHPR pathogens are found in either of the parents for a particular mating, the resulting lot of zygotes is to be destroyed. Progeny from disease-free eggs are to be retained in the quarantine facility for four months subsequent to "first-feeding". Parr samples are to be subjected to FHPR analysis after four months of active feeding. If any bacterial or viral FHPR agents are found in parr, all are to be destroyed.

Now that we have identified the conditions that would have to be met, we have to look at the facilities, equipment and expertise that will be required to get started. If we are to proceed, we will need a pressure vessel to induce the triploid condition. Since these vessels operate under high pressure, we also should consider providing the necessary expertise to operate the pressure vessel. To summarize the situation, we have: 1) a potential supply of homogametic milt at Bay d'Espoir; 2) a quarantine facility at Bay d'Espoir; 3) a certified hatchery at Port Rexton; and, 4) definition of the prerequisites to allow us to bring in unfertilized eggs. We need: 1) a pressure vessel; and, 2) additional tank space. Before we can complete this balance sheet of assets and needs, I need to know what other interests there are around the table.

Meaney We need to determine the size of the program necessary to provide adequate demonstration of the merits of this technology. Having once decided on the number of smolts required for comparative performance evaluation, we can calculate, if we know what losses to expect from triploid eggs, the number of eggs we will have to import. Will we have to fit our requirements within the 100,000 egg limit imposed by DFO or will the triploidy program be incremental to this limit?

Penney As I stated earlier, the 100,000 egg limit was defined on the basis of genetic concerns. Therefore, if we are talking triploid eggs, the requirements are likely to be incremental to the present 100,000 limit.

Pepper With all of this as background, I would like to focus on industry's perceptions of priorities. Being that Bay d'Espoir has worked with triploids and now is poised for further work, I would like to start with comments from the Bay d'Espoir Salmon Growers.

Collier We have triploid rainbow trout already and, in the context of our continued involvement with these rainbow trout, we consider salmon to be our next priority. If our present experiments with Atlantic salmon work out, demand for this species by the growers will escalate rapidly. As well, there is need for more research on Atlantic salmon. Therefore, we would place our priority on evaluations of triploid salmon performance.

Pepper Considering that you already are working with rainbow trout as your main cash flow generator, would any of the triploid salmon work preclude any of your work with rainbow trout.

Collier We would have to make sure that it does not. This will require some expansion of hatchery facilities. In fact, we likely would mount parallel endeavours with both species anyway but the scope of the activity may increase.

Freeman Regardless of who the producers are or what species they are producing, their purpose is to make money. In the context of triploid char or brood trout, if certified eggs are available, we would be interested in evaluating them. However, if either the eggs or sperm are not certified, there is an element of risk to the producer. We have to build a business on what we can get. Can we hope, realistically, to get the required all-female milt without risk to our present operations? I do not want to risk being decertified.

Pepper Do I interpret correctly that you have no immediate interest in becoming involved in a program with triploid char and brook trout because of the implications to your present operations?

Freeman I am definitely interested in small scale evaluations of the technology if I could get my hands on some fish to work with that will not jeopardize my operations. Right now, my choice would be to import certified, triploid brook trout eggs from Quebec.

Penney It seems we have the means whereby Bay d'Espoir can provide the facilities for Atlantic salmon and rainbow trout and Bob Freeman's hatchery can support an independent initiative on rainbow trout and brook trout. While Bob will have Arctic char eggs, he would lose his present certification if he were to use female sperm from Bay d'Espoir. Arctic char may have to be a sideline for the time being. All we need now to get working on three of the four species is a pressure vessel and the necessary expertise to operate it.

Pepper Have we then identified the critical elements required to implement a program this fall? What proposals are required and who will develop them? It seems there is a general need for a pressure vessel that will be available to the industry on an as required basis. Then I see the need for hatchery facilities that will be the specific property of individuals within the industry. Should there be one funding proposal that addresses all of the specific elements or should there be several individual proposals?

Meaney The important feature is the level of funding that will be required. However it is done, the proposal(s) should be linked and submitted in reference to the Dartmouth workshop proceedings document. This would help provide substantial validity within the government circles that would be involved in reviewing the proposals. This would convey that the proposals are in line with the overall needs of a technology demonstration/transfer program for triploid salmon.

Pepper Recognizing that this is a government-industry initiative, and that the pressure vessel represents a common need for Newfoundland work with triploid salmon, is it reasonable to assume that the two levels of government represented here today (i.e., Meaney and Pepper) should collaborate on the proposal for the pressure vessel and the expertise to use it? On the other hand, I cannot see government proposing specific facilities for individuals, who may or may not want to be involved in such a program, and therefore suggest that interested growers develop their own proposals, for submission under the auspices of a Newfoundland demonstration program on non-maturing salmonids.

Meaney It would be appropriate for Bob Freeman to develop the proposal for his facility at Port Rexton and Clyde Collier to identify what is required at the Bay d'Espoir Hatchery.

Brown We should not lose track of the fact that Bob Freeman's facility is certified. Therefore, if the facility

can provide sufficient fish for growout, it could provide the opportunity to evaluate growout performance at other sites such as the Great Northern Peninsula, Valley Char and Holyrood Pond. This should be considered in developing the facilities proposal(s).

Turner Our cage operation at Grand Lake is entirely fresh water. This would provide an additional factor in your program of performance evaluation.

Sutterlin We have to be careful relative to the species we include in the program. We have techniques for salmon and rainbow trout. The procedures have not been worked out for Arctic char. We have the means to get into a demonstration program with Atlantic salmon and rainbow trout. With char, we will have some research to do to figure out reliable techniques to get effective triploidy.

Yetman Do you see Holyrood Pond being involved in this program?

Pepper Given the various factors we have discussed, I feel I would have to leave this up to you to identify how and when the Holyrood Pond facility could be involved. As I see it now, you will not have access to Atlantic salmon until 1992 at the earliest. If I understand correctly, these salmon would be normal diploids. Therefore, I do not see the means whereby Holyrood Pond would be involved immediately. I will have to defer further consideration of this to you. In the mean time, I think we must establish some action items and some time frames. Can we have the proposals submitted before the end of August? Recognize that whatever program we go for, if it is to get anywhere this year, it will have to start in October.

Meaney Relative to the pressure vessel, there are legal and safety constraints inherent in the manufacture of this device. This will greatly complicate the tendering process. Hence, if there is to be any hope of having a working pressure vessel available for this October, the proposal will have to be ready within the next couple of weeks. I suggest the proposal be developed in consultation with the present Newfoundland expertise on the topic and therefore suggest Dr. Sutterlin be involved as well. Is there a general agreement from the industry that a pressure vessel is required?

Collier Agreed.
Freeman Agreed.
Turner Agreed.
Wilton Agreed.
Yetman Agreed.

Freeman What is the likely time frame for the proposed program?

Pepper For the moment, we are talking only about implementation. The duration of the program will depend on the results we get and the number of species that ultimately are included in a demonstration program. I suspect we may be looking at several years since it is quite likely we will want to make refinements to our program as we go. The first step will be the proposal for the pressure vessel. This will be pursued between the two levels of government during July. The proposals for appropriate support facilities are to be submitted to Brian Meaney or myself by the week of August 26 - 30. In the mean time, I will provide you with the minutes of this meeting as soon as possible and subsequently, will inform you as to what comes of the various proposals. Thank you for your cooperation and your contribution to this meeting.